IS 7123 (1993): Hair Oils [PCD 19: Cosmetics]

BLANK PAGE

IS 7123 : 1993
(Reaffirmed 1998)

भारतीय मानक

केश तेल — विशिष्टि

(दूसरा पुनरीक्षण)

Indian Standard

HAIR OILS — SPECIFICATION

(Second Revision)

First Reprint SEPTEMBER 2000

UDC 665·585·47

BUREAU OF INDIAN STANDARDS
MANAK BHAVAN, 9 BAHADUR SHAH ZAFAR MARG
NEW DELHI 110002

June 1993

Price Group 2

Cosmetics Sectional Committee, PCD 19

FOREWORD

This Indian Standard (Second Revision) was adopted by the Bureau of Indian Standards, after the draft finalized by the Cosmetics Sectional Committee, had been approved by the Petroleum, Coal and Related Products Division Council.

This standard was first published in 1973 when amongst others, a requirement for peroxide value of 7·5 milliequivalents/1 000 g maximum was prescribed. Vegetable oils have an inherent tendency to undergo oxidation on storage which is enhanced by direct sunlight and high temperature, thereby resulting in an increased peroxide value. At a certain stage of oxidation, the oil starts giving unpleasant rancid odour. Requirement for peroxide value was prescribed to check rancidity of hair oil. Through use of this standard, manufacturer's difficulty in complying to this requirement was realized.

Though, there is a direct correlation of peroxide value with the degree of unpleasant rancid odour of hair oil which would be tolerable to the consumer, the Committee removed the requirement of peroxide value and introduced a date up to which the product may be used to safeguard the interest of consumers, in the first revision of this standard.

Later on, it was found that, expiry date being a part of the marking clause was not complied by most of the manufacturers of hair oil and requirement of peroxide value which takes care of rancidity of hair oil had been removed from the standard in the first revision.

Lately, some of the hair oil manufacturers themselves observed that hair oil being a consumer non-durable item, requirement of peroxide value may be restored to, to check the unpleasant rancid odour of hair oil. Therefore, in this revision, a requirement for peroxide value has been included along with an expiry date as a regular requirement. Another important requirement for microbiological examination of hair oil has been included. Besides, it has been made compulsory to declare the list of critical ingredients used in hair oil on packing as well as packaging materials.

Hair oils for which therapeutic claims are made are not covered in this standard.

It is necessary that all ingredients used are such that in the concentration in which they would be present in the hair oil, are free from any harmful effects. For determining the dermatological safety of a new formulation, or of a new raw material in an old formulation, reference may be made to IS 4011 : 1982 'Methods for dermatological testing of cosmetics (*first revision*)' for prophetic testing. It shall be the responsibility of the manufacturer to satisfy himself of the dermatological safety of his formulation according to this standard before releasing the product for sale.

For the purpose of deciding whether a particular requirement of this standard is complied with, the final value, observed or calculated, expressing the result of a test or analysis, shall be rounded off in accordance with IS 2 · 1960 'Rules for rounding off numerical values (*revised*)'. The number of significant places retained in the rounded off value should be the same as that of the specified value in this standard.

AMENDMENT NO. 4 APRIL 2001
TO
IS 7123 : 1993 HAIR OILS — SPECIFICATION

(Second Revision)

(*Page* 1, *clause* 4.2.1.1 *and Amendments No.* 1 *and* 2) — Insert the following after (m) :

'n) Any other edible oil conforming to and permitted by PFA and IS 4707 (Part 1) and IS 4707 (Part 2).'

(*Page* 2, *clause* 4.2.1.3 *and Amendments No.* 1 *and* 2) — Insert the following after (n):

'p) Any other edible oil conforming to and permitted by PFA and IS 4707 (Part 1) and IS 4707 (Part 2).'

[*Page* 2, *clause* 5.2(f) *and* (g) *and Amendmend No.* 1] — Substitute the following for the existing:

'f) Best use before.....(Month and year to be declared by the manufacturer).

NOTE — This is exempted in case of pack sizes of 10 g/25 ml or less and if the shelf life of the product is more than 24 months.

'g) List of key ingredients.

NOTE — This is exempted in case of pack sizes of 30 g/60 ml or less.'

(PCD 19)

Reprography Unit, BIS, New Delhi, India

AMENDMENT NO. 5 SEPTEMBER 2006
TO
IS 7123 : 1993 HAIR OILS — SPECIFICATION

(Second Revision)

(*Page* 2, *clause* 4.6) – Delete.

(*Page* 2, *clause* 4.7) – Delete.

(*Page* 2, *clause* 5.1) – Substitute the following for the existing.

'The hair oil shall be packed in a suitable well closed container, not exceeding pack size of 1 kg or 1 litre, and the container should not have deleterious effect on the product.'

[*Page* 2, *clause* 5.2(g) (*see also Amendment No.* 4)] – Substitute the following for the existing:

'*List all ingredients* — Ingredients present at greater than 1 percent shall be listed in descending order of mass at the time they are added, followed by those in concentration of less than or equal to 1 percent in any order Colouring agents may be listed in any order after the other ingredients

NOTES

1 This is exempted in case of pack sizes of 30 g, 60 ml or less

2 Composition percentage of mineral oil as well as vegetable oil shall be declared in Type 3 Hair Oil.'

(PCD 19)

Reprography Unit, BIS, New Delhi, India

AMENDMENT NO. 5 SEPTEMBER 2006
TO
IS 7123 : 1993 HAIR OILS — SPECIFICATION

(Second Revision)

(*Page* 2, *clause* **4.6**) – Delete.

(*Page* 2, *clause* **4.7**) – Delete

(*Page* 2, *clause* **5.1**) – Substitute the following for the existing:

'The hair oil shall be packed in a suitable well closed container, not exceeding pack size of 1 kg or 1 litre, and the container should not have deleterious effect on the product '

[*Page* 2, *clause* **5.2**(g) (*see also Amendment No.* 4)] – Substitute the following for the existing:

'*List all ingredients* — Ingredients present at greater than 1 percent shall be listed in descending order of mass at the time they are added, followed by those in concentration of less than or equal to 1 percent in any order Colouring agents may be listed in any order after the other ingredients

NOTES

1 This is exempted in case of pack sizes of 30 g/60 ml or less.

2 Composition percentage of mineral oil as well as vegetable oil shall be declared in Type 3 Hair Oil '

(PCD 19)

Reprography Unit, BIS, New Delhi, India

AMENDMENT NO. 1 DECEMBER 1995
TO
IS 7123 : 1993 HAIR OILS — SPECIFICATION

(Second Revision)

(*Page* 1, *clause* 2.1) — Insert the following:

'IS 546 : 1976 Mustard oil (*second revision*)' between 'IS 543 : 1975' and 'IS 1070 : 1992'.

(*Page* 1, *clause* 4.2.1.1) — Add the following after 'h':

'j) Mustard oil conforming to IS 546 : 1975'

(*Page* 2, *clause* 4.2.1.3) — Add the following after 'j':

'k) Mustard oil conforming to IS 546 : 1975'

(*Page* 2, *clause* 4.4) — Substitute '1 000 micro organisms per gram' *for* '100 micro organisms per gram'.

(*Page* 2, *clause* 4.5) — Substitute '10.0 milliequivalents/1 000 g' *for* '7.5 milliequivalents/1 000g'.

[*Page* 2, *clause* 5.2(f)] — Substitute the following for the existing:

'Best used before _____ (month and year)'.

Reprography Unit, BIS, New Delhi, India

AMENDMENT NO. 2 DECEMBER 1997
TO
IS 7123 : 1993 HAIR OILS — SPECIFICATION

(Second Revision)

[*Page* 1, *clause* 2.1 (*see also Amendment No.* 1)] — Insert the following:

a) 'IS 3448 : 1984 Ricebran oil (*second revision*)' between 'IS 1070 : 1992' and 'IS 3491 : 1965', and

b) 'IS 5637 : 1970 Watermelon seed oil' between 'IS 4707 (Part 2) : 1993' and 'IS 7299 : 1974'.

[*Page* 1, *clause* 4.2.1.1 (*see also Amendment No.* 1)] — Insert the following after (j):

'k) Ricebran oil conforming to IS 3448 : 1984

m) Watermelon seed oil conforming to IS 5637 : 1970'.

(*Page* 2, *clause* 4.2.1.3 (*see also Amendment No.* 1) — Insert the following after 'k' :

'm) Ricebran oil conforming to IS 3448 : 1984

n) Watermelon seed oil conforming to IS 5637 : 1970'.

(PCD 19)

Reprography Unit, BIS, New Delhi, India

AMENDMENT NO. 3 OCTOBER 1998
TO
IS 7123 : 1993 HAIR OILS — SPECIFICATION

(Second Revision)

(*Foreword, para* 7) — Insert the following after para 7:

'A scheme for labelling environment friendly products known as ECO Mark has been introduced at the instance of the Ministry of Environment and Forests (MEF), Government of India. The ECO Mark is being administered by the *Bureau of Indian Standards Act*, 1986 as per the Resolution No. 71 dated 21 February 1991 and No.768 dated 24 August 1992 published in the Gazette of the Government of India. For a product to be eligible for marking with ECO logo, it shall also carry the Standard Mark of BIS besides meeting additional environment friendly requirements. For this purpose, the Standard Mark of BIS would be a single mark being a combination of the BIS monogram ISI and the ECO logo. Requirements for ECO friendliness will be additional, manufacturing units will be free to opt for Standard Mark alone also.

This amendment is based on the Gazette Notification No. 170 dated 18 May 1996 for hair oils as environment friendly products published in the Gazette of the Government of India. This amendment is, therefore, being issued to this standard to include environment friendly requirements for hair oil.'

(*Page* 1, *clause* 2.1) — Insert the following after IS 3958 : 1984:

'IS 4011 : 1997 Methods of test for safety evaluation of cosmetics (*second revision*)'

(*Page* 2, *clause* 4.5) — Insert the following clauses after 4.5 and renumber the subsequent clauses accordingly:

4.6 Additional Requirements for ECO Mark

4.6.1 *General Requirements*

4.6.1.1 The product shall conform to the requirements for quality, safety and performance prescribed under 4.1 to 4.5.

4.6.1.2 All the ingredients that go into formulation of cosmetics shall comply with the provisions of IS 4707 (Part 1) : 1988 'Classification of cosmetic raw materials and adjuncts: Part 1 Dyes, colours and pigments (*first revision*)' and IS 4707 (Part 2) : 1993 'Classification of cosmetic raw materials and adjuncts: Part 2 List of raw materials generally not recognized as safe (*first revision*)'.

Indian Standard

HAIR OILS — SPECIFICATION

(Second Revision)

1 SCOPE

1.1 This standard prescribes the requirements for hair oils and other oil-based cosmetic preparations for the hair. The latter include hair tonics and hair oil concentrates.

1.1.1 This standard does not covers enfluerage type of hair oils, hair creams, brilliantines, pomades and preparations sold under the name of hair darkeners.

1.1.2 Hair oils for which therapeutic claims are made are not covered in this standard.

2 REFERENCES

2.1 The following Indian Standards are necessary adjuncts to this standard

IS No.	*Title*
543 : 1975	Cottonseed oil (*second revision*)
1070 : 1992	Reagent grade water (*third revision*)
3491 : 1965	Safflower oil
3958 : 1984	Methods of sampling cosmetics (*first revision*)
4276 : 1977	Soyabean oil (*first revision*)
4277 : 1975	Sunflower oil (*first revision*)
4707 (Part 1) : 1988	Classification for cosmetic raw materials and adjuncts: Part 1 Dyes, pigments and colours (*first revision*)
4707 (Part 2) : 1993	Classification for cosmetic raw materials and adjuncts: Part 2 List of raw materials generally not recognized as safe for use in cosmetics (*first revision*)
7299 : 1974	Mineral oil for cosmetics industry (*first revision*)
11375 : 1985	*Groundnut oil for cosmetic* industry
11376 : 1985	Sesame oil for cosmetic industry
11470 : 1985	Coconut oil for cosmetic industry
11486 : 1985	Castor oil for cosmetic industry

The above mentioned standards contain provisions which, through reference in this text, constitute provisions of this standard. At the time of publication, the editions indicated were valid. All standards are subject to revision, and parties to agreements based on this standard are encouraged to investigate the possibility of applying the most recent editions of the standards indicated above.

3 TYPES

3.1 There shall be three types of hair oils, namely:

Type 1 based on vegetable oil or oils,

Type 2 based on mineral oil, and

Type 3 based on a mixture of vegetable oils(s) (raw and/or refined) and mineral oil.

4 REQUIREMENTS

4.1 Description

The hair oil shall be colourless or coloured, with or without perfume. It shall be free from any sediment and suspended matter at 27°C and unpleasant rancid odour. It shall contain suitable antioxidants, if necessary, to prevent it from developing unpleasant rancid odour till the time of its actual use.

4.2 Ingredients

4.2.1 *Base Oil*

4.2.1.1 *For type 1*

The oil or oils used as the base shall be of the quality specified below except for the requirement of colour when used in the formulation of coloured hair oil.

a) Castor oil conforming to IS 11486 : 1985,

b) Coconut oil conforming to IS 11470 : 1985,

c) Groundnut oil conforming to IS 11375 : 1985,

d) Sesame oil conforming to IS 11376 : 1985,

e) Cottonseed oil conforming to IS 543 : 1975,

f) Sunflower oil conforming to IS 4277 : 1975,

g) Safflower oil conforming to IS 3491 : 1965, and

h) Soyabean oil conforming to IS 4276 : 1977.

Indian Standard

HAIR OILS — SPECIFICATION

(Second Revision)

1 SCOPE

1.1 This standard prescribes the requirements for hair oils and other oil-based cosmetic preparations for the hair. The latter include hair tonics and hair oil concentrates.

1.1.1 This standard does not covers enfluerage type of hair oils, hair creams, brilliantines, pomades and preparations sold under the name of hair darkeners.

1.1.2 Hair oils for which therapeutic claims are made are not covered in this standard.

2 REFERENCES

2.1 The following Indian Standards are necessary adjuncts to this standard:

IS No.	*Title*
543 : 1975	Cottonseed oil (*second revision*)
1070 : 1992	Reagent grade water (*third revision*)
3491 : 1965	Safflower oil
3958 : 1984	Methods of sampling cosmetics (*first revision*)
4276 : 1977	Soyabean oil (*first revision*)
4277 : 1975	Sunflower oil (*first revision*)
4707 (Part 1) : 1988	Classification for cosmetic raw materials and adjuncts: Part 1 Dyes, pigments and colours (*first revision*)
4707 (Part 2) : 1993	Classification for cosmetic raw materials and adjuncts: Part 2 List of raw materials generally not recognized as safe for use in cosmetics (*first revision*)
7299 : 1974	Mineral oil for cosmetics industry (*first revision*)
11375 : 1985	Groundnut oil for cosmetic industry
11376 : 1985	Sesame oil for cosmetic industry
11470 : 1985	Coconut oil for cosmetic industry
11486 : 1985	Castor oil for cosmetic industry

The above mentioned standards contain provisions which, through reference in this text, constitute provisions of this standard. At the time of publication, the editions indicated were valid. All standards are subject to revision, and parties to agreements based on this standard are encouraged to investigate the possibility of applying the most recent editions of the standards indicated above.

3 TYPES

3.1 There shall be three types of hair oils, namely:

Type 1 based on vegetable oil or oils,

Type 2 based on mineral oil, and

Type 3 based on a mixture of vegetable oils(s) (raw and/or refined) and mineral oil.

4 REQUIREMENTS

4.1 Description

The hair oil shall be colourless or coloured, with or without perfume. It shall be free from any sediment and suspended matter at 27°C and unpleasant rancid odour. It shall contain suitable antioxidants, if necessary, to prevent it from developing unpleasant rancid odour till the time of its actual use.

4.2 Ingredients

4.2.1 *Base Oil*

4.2.1.1 *For type 1*

The oil or oils used as the base shall be of the quality specified below except for the requirement of colour when used in the formulation of coloured hair oil.

a) Castor oil conforming to IS 11486 : 1985,

b) Coconut oil conforming to IS 11470 : 1985,

c) Groundnut oil conforming to IS 11375 : 1985,

d) Sesame oil conforming to IS 11376 : 1985,

e) Cottonseed oil conforming to IS 543 : 1975,

f) Sunflower oil conforming to IS 4277 : 1975,

g) Safflower oil conforming to IS 3491 : 1965, and

h) Soyabean oil conforming to IS 4276 : 1977.

Indian Standard

HAIR OILS — SPECIFICATION

(Second Revision)

1 SCOPE

1.1 This standard prescribes the requirements for hair oils and other oil-based cosmetic preparations for the hair. The latter include hair tonics and hair oil concentrates.

1.1.1 This standard does not covers enfluerage type of hair oils, hair creams, brilliantines, pomades and preparations sold under the name of hair darkeners.

1.1.2 Hair oils for which therapeutic claims are made are not covered in this standard.

2 REFERENCES

2.1 The following Indian Standards are necessary adjuncts to this standard

IS No.	*Title*
543 : 1975	Cottonseed oil (*second revision*)
1070 : 1992	Reagent grade water (*third revision*)
3491 : 1965	Safflower oil
3958 : 1984	Methods of sampling cosmetics (*first revision*)
4276 : 1977	Soyabean oil (*first revision*)
4277 : 1975	Sunflower oil (*first revision*)
4707 (Part 1) : 1988	Classification for cosmetic raw materials and adjuncts: Part 1 Dyes, pigments and colours (*first revision*)
4707 (Part 2) : 1993	Classification for cosmetic raw materials and adjuncts: Part 2 List of raw materials generally not recognized as safe for use in cosmetics (*first revision*)
7299 : 1974	Mineral oil for cosmetics industry (*first revision*)
11375 : 1985	Groundnut oil for cosmetic industry
11376 : 1985	Sesame oil for cosmetic industry
11470 : 1985	Coconut oil for cosmetic industry
11486 : 1985	Castor oil for cosmetic industry

The above mentioned standards contain provisions which, through reference in this text, constitute provisions of this standard. At the time of publication, the editions indicated were valid. All standards are subject to revision, and parties to agreements based on this standard are encouraged to investigate the possibility of applying the most recent editions of the standards indicated above

3 TYPES

3.1 There shall be three types of hair oils, namely

Type 1 based on vegetable oil or oils,

Type 2 based on mineral oil, and

Type 3 based on a mixture of vegetable oils(s) (raw and/or refined) and mineral oil

4 REQUIREMENTS

4.1 Description

The hair oil shall be colourless or coloured, with or without perfume. It shall be free from any sediment and suspended matter at 27°C and unpleasant rancid odour. It shall contain suitable antioxidants, if necessary, to prevent it from developing unpleasant rancid odour till the time of its actual use

4.2 Ingredients

4.2.1 *Base Oil*

4.2.1.1 *For type 1*

The oil or oils used as the base shall be of the quality specified below except for the requirement of colour when used in the formulation of coloured hair oil

a) Castor oil conforming to IS 11486 : 1985,

b) Coconut oil conforming to IS 11470 : 1985,

c) Groundnut oil conforming to IS 11375 : 1985,

d) Sesame oil conforming to IS 11376 : 1985,

e) Cottonseed oil conforming to IS 543 : 1975,

f) Sunflower oil conforming to IS 4277 : 1975,

g) Safflower oil conforming to IS 3491 : 1965, and

h) Soyabean oil conforming to IS 4276 : 1977.

4.2.1.2 *For type 2*

The base oil shall conform to IS 7299 : 1974.

4.2.1.3 *For type 3*

The oil or oils used as the base shall be of the quality specified below except for the requirement of colour when used in the formulation of coloured hair oil.

a) Castor oil conforming to IS 11486 : 1985,

b) Coconut oil conforming to IS 11470 : 1985,

c) Groundnut oil conforming to IS 11375 : 1985,

d) Sesame oil conforming to IS 11376 : 1985,

e) Cottonseed oil conforming to IS 543 : 1975,

f) Sunflower oil conforming to IS 4277 : 1975,

g) Safflower oil conforming to IS 3491 : 1965,

h) Soyabean oil conforming to IS 4276 : 1977, and

j) Mineral oil conforming to IS 7299 : 1974.

4.2.2 *Dyes and Colours*

If dyes and colours are to be added, these shall be those included in Schedule Q of Drugs and Cosmetics Rules and shall conform to the requirements prescribed in IS 4707 (Part 1) : 1988.

4.2.3 *Other Additives*

Other additives shall conform to the requirements prescribed in IS 4707 (Part 2) : 1993.

4.3 Acid Value

The acid value of Type 1 and Type 3 hair oils when tested as prescribed in A-2.1 shall be not more than 1·0. The hair oil of Type 2 shall pass the test prescribed in A-2.2.

4.4 Microbiological Examination

The material when tested as prescribed in A-3 shall contain not more than 100 micro-organisms per gram.

4.5 Peroxide Value

The material when tested as prescribed in A-4 shall have peroxide value not exceeding 7·5 milliequivalents/1 000 g.

4.6 Expiry Date

Should be declared by the manufacturer. Date up to which the product may be used should be mentioned on the packing material. The product may be used maximum up to three years from the date of its manufacture.

4.7 Labelling

List of critical ingredients should be mentioned on the packing material.

5 PACKING AND MARKING

5.1 Packing

The hair oil shall be packed in suitable well-closed containers which do not have deleterious effect on the product.

5.2 Marking

The containers shall be legibly marked with the following information:

a) Manufacturer's name and recognized trade-mark, if any;

b) Indication of the source of manufacture and type of the material;

c) Net contents of the material in ml or g;

d) Batch number, in code or otherwise, to enable the lot of material to be traced back from records;

e) Store in cool place protected from sunlight;

f) Expiry date; and

g) List of critical ingredients.

5.2.1 The containers may also be marked with the Standard Mark.

6 SAMPLING

6.1 Representative samples of the material shall be drawn as prescribed in IS 3958 : 1984.

6.2 Tests for all the requirements shall be carried out on the composite sample.

6.3 The material shall be taken to have conformed to this specification if the composite sample passes all the tests.

ANNEX A

(*Clauses* 4.3, 4.4 *and* 4.5)

METHODS OF TEST FOR HAIR OILS

A-1 QUALITY OF REAGENTS

A-1.1 Unless specified otherwise, pure chemicals and distilled water (*see* IS 1070 : 1992) shall be used in tests.

NOTE — 'Pure chemicals' shall mean chemicals that do not contain impurities which affect the results of analysis

A-2 DETERMINATION OF ACID VALUE

A-2.1 For Type 1 and Type 3

A-2.1.1 *Reagents*

A-2.1.1.1 *Ethyl alcohol*

95 percent by volume, neutralized to mixed indicator solution (**A-2.1.1.2**).

A-2.1.1.2 *Mixed indicator solution*

Dissolve 1 g of phenolphthalein in 100 ml of ethyl alcohol and add to it 1 ml of 0·1 percent solution of methylene blue in water.

A-2.1.1.3 *Standard aqueous potassium hydroxide or sodium hydroxide solution* 0·1 N.

A-2.1.2 *Procedure*

Weigh accurately a suitable quantity of the oil in a 200-ml conical flask. The mass of the oil taken shall be such that the volume of alkali required for the titration does not exceed 10 ml. Add 50 ml of hot ethyl alcohol and 1 ml of the mixed indicator solution. Boil the mixture for about 5 minutes and titrate while hot with standard alkali solution, shaking vigorously during titration.

A-2.1.3 *Calculation*

$$\text{Acid value} = \frac{56{\cdot}1\ V\ N}{M}$$

where

V = volume in ml of standard alkali solution used;

N = normality of standard alkali solution; and

M = mass in g of the material taken for the test.

A-2.2 For Type 2

A-2.2.1 *Procedure*

Shake 20 g of the material with an equal amount of hot distilled water. Test the aqueous portion with blue litmus paper.

A-2.2.1.1 The material shall be taken to have passed the test if the litmus does not change colour.

A-3 MICROBIOLOGICAL EXAMINATION

A-3.0 Outline of the Method

The test consists of plating a known mass of the sample on two selected culture media specifically suitable for the growth of bacteria and fungi and incubating them for a specified period to permit the development of visual colonies for counting.

A-3.1 Apparatus

A-3.1.1 *Tubes*, of resistant glass, provided with closely fitting metal caps.

A-3.1.2 *Autoclaves*, of suitable size

They shall keep uniform temperature within the chamber up to and including the sterilizing temperature of 120°C. They shall be equipped with an accurate thermometer, located so as to register the minimum temperature within the sterilizing chamber, a pressure gauge and properly adjusted safety valves.

A-3.1.3 *Petri Dishes*

Of 100 mm diameter and 15 mm depth. The bottom of the dishes shall be free from bubbles and scratches and shall be flat so that the medium is of uniform thickness throughout the plate

A-3.1.4 *Colony Counter*

An approved counting aid, such as Quebec colony counter. If such a counter is not available, counting may be done with a lens giving a magnification of 1·5 diameter. In order to ensure uniformity of conditions during counting illumination equivalent to that provided by the Quebec colony counter shall be employed.

A-3.2 Media

A-3.2.1 *Nutrient Agar Medium*

Dissolve 5 g of yeast extract (or meat extract), 5 g of sodium chloride and 10 g of peptone in 1 000 ml of distilled water contained in a 2-litre beaker by heating on a water-bath. Add 25 g of powdered agar and continue boiling until the agar is completely dissolved. Adjust the *p*H to 7·4 with sodium hydroxide solution using *p*H meter or comparator. Filter while hot through lint cloth placed in a funnel and dispense into

tubes in 20 ml quantities. Filter only if necessary. Close the tubes with metal caps or cotton plugs and sterilize in an autoclave at 121°C and 1·05 kgf/cm² pressure for 20 minutes. After autoclaving, store the tubes in a refrigerator and use them within 3 weeks.

A-3.2.2 *Sabouraud Agar Medium*

Dissolve 10 g of peptone and 40 g of glucose in 1 000 ml of distilled water contained in a 2-litre conical flask by heating in water-bath. Add 25 g of powdered agar and continue boiling until the agar is completely dissolved. *p*H need not be adjusted (it automatically comes to 5·4). Filter while hot through lint cloth placed in a funnel and dispense into tubes in 20-ml quantities. Filter only, if necessary. Close the tubes with metal caps or cotton plugs and sterilize in an autoclave at 121°C and 1·05 kgf/cm² pressure for 15 minutes. After autoclaving, store the tubes in a refrigerator and use them within 3 weeks.

A-3.3 Sterilization of Apparatus

A-3.3.1 *Tubes*

These shall be sterilized in the autoclave at 121°C and 1·05 kgf/cm² pressure for 20 minutes or in a hot air oven at 160°C for one hour.

A-3.3.2 *Petri Dishes*

These shall be packed in drums and sterilized in the autoclave at 121°C and 1·05 kgf/cm² pressure for 20 minutes or individually wrapped in kraft paper and sterilized in a hot air oven at 160°C for one hour.

A-3.3.3 *Pipettes*

These shall be placed in pipettes cone (of copper, stainless steel, or aluminium) after plugging the broader end with cotton and sterilized in a autoclave at 121°C and 1·05 kgf/cm² pressure for 20 minutes or in a hot air oven at 160°C for one hour.

A-3.4 Procedure

A-3.4.1 Melt sufficient number of nutrient agar tubes and sabouraud agar tubes in a water-bath and transfer while hot into a constant temperature water-bath maintained at 48 ± 2°C.

A-3.4.2 Weigh and transfer aseptically four 0·5 g portions of the sample to four melted nutrient agar tubes, and four 0·5 g portions to four sabouraud agar tubes. Shake the tubes to mix the contents thoroughly and pour into sterile petri dishes. Incubate the nutrient agar tubes at 37 ± 0·5 C for 48 h and the sabouraud agar tubes at 20 to 25°C for 7 days

A-3.5 Determine the average number of colonies per gram of the sample on nutrient agar tubes, as well as, the average number of colonies per gram of sample on sabouraud agar tubes. The mean of the two average number shall be taken as the number of micro-organisms per gram of the samples.

A-4 TEST FOR PEROXIDE VALUE

A-4.1 Reagents

A-4.1.1 *Glacial Acetic Acid*

A-4.1.2 *Chloroform*

A-4.1.3 *Potassium Iodide Solution*, saturated, freshly prepared

A-4.1.4 *Standard Sodium Thiosulphate Solution*, 0·01 N, freshly standardized.

A-4.1.5 *Starch Indicator Solution*

Triturate 5 g of starch and 0·01 g of mercuric iodide with 30 ml of cold water and slowly pour it with stirring into one litre of boiling water Boil for three minutes. Allow to cool and decant off the supernatant clear liquid

A-4.2 Procedure

Weigh accurately about 5 g of the material in a 250-ml glass stoppered conical flask and dissolve by shaking in 30 ml of a mixed solvent containing 3 parts by volume of glacial acetic acid and 2 parts by volume of chloroform. Add 0·5 ml of saturated potassium iodide solution, allow the solution to stand for exactly 1 minute with occasional shaking, then add 30 ml of water and titrate with standard sodium thiosulphate solution. Add the thiosulphate solution until the colour of the titrated solution becomes light yellow. Then add 1 ml of starch indicator solution and continue the titration till the disappearance of the blue colour

Carry out a blank determination without using the sample.

A-4.2.1 *Calculation*

$$\text{Peroxide value milliequivalents 1 000 g} = \frac{1\,000\,(V_1 - V_2)\,N}{M}$$

where

V_1 = volume in ml of standard sodium thiosulphate solution required with the sample,

V_2 = volume in ml of standard sodium thiosulphate solution required with the blank,

N = normality of standard sodium thiosulphate solution and

M = mass in g of the sample taken for the test.

Bureau of Indian Standards

BIS is a statutory institution established under the *Bureau of Indian Standards Act*, 1986 to promote harmonious development of the activities of standardization, marking and quality certification of goods and attending to connected matters in the country.

Review of Indian Standards

Amendments are issued to standards as the need arises on the basis of comments. Standards are also reviewed periodically; a standard along with amendments is reaffirmed when such review indicates that no changes are needed; if the review indicates that changes are needed, it is taken up for revision. Users of Indian Standards should ascertain that they are in possession of the latest amendments or edition by referring to the latest issue of 'BIS Handbook' and 'Standards Monthly Additions'.

This Indian Standard has been developed from Doc. No. PCD 19 (1141)

Amendments Issued Since Publication

Amend No.	Date of Issue	Text Affected

BUREAU OF INDIAN STANDARDS

Headquarters:

Manak Bhavan, 9 Bahadur Shah Zafar Marg, New Delhi 110002
Telephones: 323 01 31, 323 33 75, 323 94 02

Telegrams: Manaksanstha
(Common to all offices)

Regional Offices:		Telephone
Central	: Manak Bhavan, 9 Bahadur Shah Zafar Marg NEW DELHI 110002	323 76 17, 323 38 41
Eastern	: 1/14 C.I.T. Scheme VII M, V.I.P. Road, Maniktola CALCUTTA 700054	337 84 99, 337 85 61 337 86 26, 337 91 20
Northern	: SCO 335-336, Sector 34-A, CHANDIGARH 160022	60 38 43 60 20 25
Southern	: C.I.T. Campus, IV Cross Road, CHENNAI 600113	235 02 16, 235 04 42 235 15 19, 235 23 15
Western	: Manakalaya, E9 MIDC, Marol, Andheri (East) MUMBAI 400093	832 92 95, 832 78 58 832 78 91, 832 78 92
Branches	: AHMADABAD. BANGALORE. BHOPAL. BHUBANESHWAR COIMBATORE. FARIDABAD. GHAZIABAD. GUWAHATI. HYDERABAD. JAIPUR. KANPUR. LUCKNOW. NAGPUR PATNA. PUNE. THIRUVANANTHAPURAM.	

Printed by Reprography Unit, BIS, New Delhi